

Restriction Digest

Requirements:

- Plasmid DNA or PCR product
- Restriction enzymes and buffers (produced by Takara Bio®)
- Nuclease-free 0.2mL PCR tubes
- Water bath at 37°C

Protocol:

1. Prepare reaction systems in a 0.2mL PCR tube according to the following table:

Components	Volume
Plasmid or PCR product	14μL
Restriction enzyme I	2μL
Restriction enzyme II	2μL
Buffer (10×)	2μL

Choose the buffer according to the following table:

	EcoR I	Xba I	Spe I	Pst I
EcoR I	H	M	H	H
Xba I	M	M+BSA	M	M
Spe I	H	M	M	H
Pst I	H	M	H	H

2. Incubate in the water bath at 37°C for 6-8 hours.